Fla. Dept. Agric. & Consumer Services
Division of Plant Industry

The Peanut Pod Nematode, *Ditylenchus africanus*[1]

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INTRODUCTION: The peanut pod nematode, *Ditylenchus africanus* Wendt, Swart, Vrain and Webster, 1995, was first found in hulls and seeds of peanut (*Arachis hypogaea* L.) in South Africa in 1987. Nematode-infected pods of the cultivar Sellie showed a black discoloration resembling black hull or black pod rot disease caused by the fungus *Chalara elegans* Nagraj and Kendrick (Figs. 1A, B) (Jones and De Waele 1988). On the basis of a comparative morphometrical and morphological study, the nematodes isolated from the peanut pods were first identified as *Ditylenchus destructor* Thorne, 1945, the potato rot nematode (De Waele *et al.* 1989).

Fig. 1. Peanut pods and seeds infected by *Ditylenchus africanus*. A) initial phase of the infection. Note dark brown tissues at the connection point between the pod base and the peg (arrows). Seeds become discolored (arrows). B) Advanced phase of infection. Note longitudinal stripes of black or brown tissue over a side of the pods (arrows) and expanding in large dark areas. Discolored and shrunken infected seeds (arrows) have flaccid seed coat marked by darked veins and can germinate prematurely (PS). Pod symptoms caused by the nematode can be confused with those of black hull or black pod rot induced by the fungus *Chalara elegans*.

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Until then, *D. destructor* was known mainly as an important pest of potato tubers and bulbs of flowers in temperate regions (localized areas in the United States, many parts of Europe, and the former Soviet Union). Experiments showed that all potato (*Solanum tuberosum* L.) cultivars tested were poor hosts of the South African population and no damage was caused to the potato tubers (De Waele *et al.* 1991), and that the optimum temperature for development of the population was 28 °C (82°F) (De Waele and Wilken 1990). Therefore, the South African population was considered a different race and ecotype. A few years later, based on differences of morphology and restriction fragment length polymorphisms (RFLPs) of ribosomal DNA (rDNA), the South African population of *D. destructor* (isolated from peanuts) was considered to be a new species and described as *D. africanus* (Wendt *et al.* 1995).

MORPHOLOGICAL CHARACTERISTICS: Female and male length of *D. africanus* is 550-1150 µm. Width 20-30 µm. Stylet delicate, 8-10 µm long, with distinct, separated, backwards sloping knobs. Head flattened. Median bulb with crescentic valves, basal bulb slightly overlapping intestine. Lateral field with 6-15 incisures. Postvulval uterine sac 1.5-3.7 times vulval body diameter. Tail elongate-conoid, tapering gradually to a finely rounded tip. Spicule 15-22 µm long, bursa enveloping 48-66 % of tail. Females and males occur in same numbers. Selected morphological characters of diagnostic significance for the separation of *D. africanus* from the related plant-parasitic species of the genus, *D. destructor* and *D. dipsaci* (Kühn, 1857) Filipjev, 1936 are listed below. *Ditylenchus africanus* differs from *D. destructor* mainly in the stylet length (8-10 µm vs. 10-14 µm) and spicule length (15.2-22 µm vs. 24-27 µm). It differs from *D. dipsaci* in the number of incisures in the lateral fields (6-15 vs. 4) (De Waele *et al.* 1989; Wendt *et al.* 1995). The morphological differences between *D. africanus* and *D. destructor* should be confirmed by examining more populations of *D. africanus*.

HOST RANGE AND DISTRIBUTION: The peanut pod nematode can survive, albeit in low numbers, without causing damage, on a variety of crops: alfalfa (*Medicago sativa* L.), corn (*Zea mays* L.), cotton (*Gossypium hirsutum* L.), cowpea (*Vigna unguiculata* (L.) Walp.), drybean (*Phaseolus vulgaris* L.), grain sorghum (*Sorghum bicolor* Moench), lupin (*Lupinus albus* L.), pea (*Pisum sativum* L.), potatoes (De Waele *et al.* 1991), soybean (*Glycine max* (L.) Merr.), sunflower (*Helianthus annuus* L.), tobacco (*Nicotiana tabacum* L.), and wheat (*Triticum aestivum L.)* (Basson *et al.* 1990). It also can survive on weeds which are common in peanut fields in South Africa: cocklebur (*Xanthium strumarium* L.), feathertop chloris (*Chloris virgata* Sw.), goose grass (*Eleusine indica* (L.) Gaertn.), khaki weed (*Tagetes minuta* L.), jimson weed (*Datura stramonium* L.), purple nutsedge (*Cyperus rotundus* L.), and white goosefoot (*Chenopodium album* L.) (De Waele *et al.* 1990). *Ditylenchus africanus* has also been observed feeding and reproducing on the hyphae of common plant pathogenic fungi: *Aspergillus parasiticus* Speare, *Botrytis cinerea* Pers.:Fr., *Fusarium oxysporum* Schlechtend.:Fr., *Fusarium solani* (Mart.) Sacc., *Macrophomina phaseolina* (Tassi) Goidanich, *Penicillium* spec. (Fig. 2; unpublished results), *Rhizoctonia solani* Kuehn, *Sclerotium rolfsii* Sacc., and on the common nematode-trapping fungus, *Arthrobotrys* sp. (Swart and Jones 1994). *Aspergillus flavus* Link:Fr. and *Aspergillus niger* Tiegn. were non-hosts (unpublished results).

The peanut pod nematode has been found in all the major peanut production areas of South Africa: 73 % of 877 seed samples examined were infected with *D. africanus* (De Waele *et al.* 1989). Its widespread distribution in South Africa suggests that it may be present in other southern African countries, especially those neighbouring South Africa. Peanut pods showing symptoms typical of *D. africanus* infection have been reported from Mozambique (Chirruco, INIA personal communication), and from the Mzuzu area of Malawi and Nkayi area of Congo (Swanevelder, ARC-GCI personal communication). In 1995, peanut seeds infected with a *Ditylenchus* spec., originating from the Pacific, were intercepted in India (Sharma, ICRISAT, personal communication). The nematode remained unnoticed in South Africa for so long probably because the symptoms it causes are very similar to those of black hull or black pod rot disease, and the nematodes themselves have a weak stylet and resemble harmless fungivorous nematodes.

BIOLOGY, CULTURING AND EXTRACTION: Temperature has a great influence on egg production, hatching and the length of the life cycle of *D. africanus* (De Waele and Wilken 1990). The optimum temperature for development of the nematode is 28 °C (82°F) (most eggs are produced; egg viability is higher than 90 % and the life-cycle (from egg to egg) is 6 - 7 days). This short life cycle enables the peanut pod nematode to build up extremely high population densities during the growing season. Inoculation of peanut seedlings with 500 nematodes per plant resulted in populations of about 150,000 nematodes per 5 g fresh hulls or seeds after 18 weeks (Bolton *et al.* 1990).

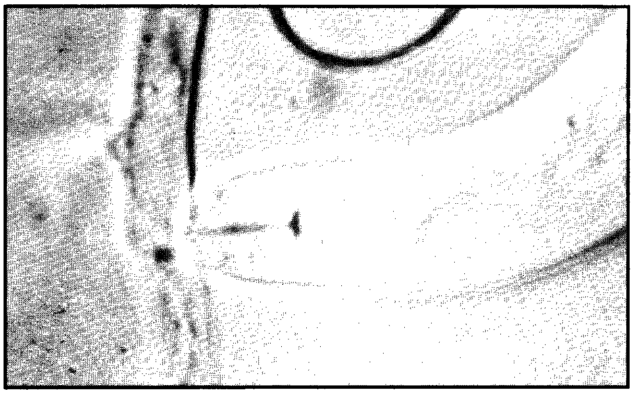

Fig. 2. *Ditylenchus africanus* feeding on a hypha of *Penicillium* species.

Mass cultures of *D. africanus* can easily be established on peanut callus tissue initiated from peanut leaves. Inoculation of callus tissue with 50 nematodes resulted in a 600-fold increase at five weeks after inoculation (Van der Walt and De Waele 1989).

Ditylenchus africanus is best extracted from hulls and seeds of peanut by soaking the tissues in shallow water in petri dishes for 24 hrs. at room temperature (Bolton *et al.* 1990).

SYMPTOMS, HISTOPATHOLOGY AND DAMAGE: Soon after the peg has entered the soil and pod formation has been initiated, the nematodes enter the plant tissues at the base of the pod near the point of connection with the peg (Jones and DeWaele 1990). The first symptom is the appearance of dark brown tissues with a corky appearance at the pod base where the peg joins the pod. The pod (Fig. 1A) may break off during harvesting. From the base of the pod, the nematodes penetrate the outer layers of the hull, feeding on the parenchyma cells surrounding the vascular tissues, causing cell wall breakage and cell malformation (Fig. 3). In advanced infections, cells collapse and the formation of tunnels facilitates nematode migration (Venter *et al.* 1995). This behavior causes the characteristic pod symptoms and their typical development (Figs. 1A, B): dark brown tissue discoloration at the base of the pod followed by the appearance of dark brown to black longitudinal stripes extending first over one side of the pod, then over both sides until the entire pod surface is discolored. Infected hulls lack the luster of healthy pods and appear dead. The severity of the pod symptoms is highly correlated with the number of nematodes in the hulls (Venter *et al.* 1991).

In immature pods, the nematodes move across the fibrous regions of the mesocarp into the endocarp of the hull. In mature pods, however, the fibrous mesocarp of the hull is lignified and, apparently, acts as a barrier against penetration of the inner hull tissues. From the endocarp of the hull, the nematodes enter the seed through the micropyle and invade the seed coat (testa), which may be entirely destroyed (Venter *et al.* 1995). Infected seeds are shrunken and the micropyles dark brown to black. The seed coat is flaccid, has darker veins ("blemished") (Figs. 1A, B) and is easily removed. The inner layer of the seed coat has a distinct yellow discoloration. Often the chemical compounds which inhibit seed germination are inactivated and as a result the seed may initiate growth of the hypocotyl ("unsound"). In severe infections, the weakened pod may split open and second-generation seedlings may sprout around the mother plant. The nematodes do not appear to penetrate the cotyledons, but do infect the embryo (Fig. 4). Infected embryos are usually olive green to brown instead of having a normal colorless to yellow appearance. The symptoms caused by *D. africanus* are similar to those caused by *Aphelenchoides arachidis* Bos, the groundnut testa nematode, which has been found in peanut seeds in Nigeria (Bridge *et al.* 1977).

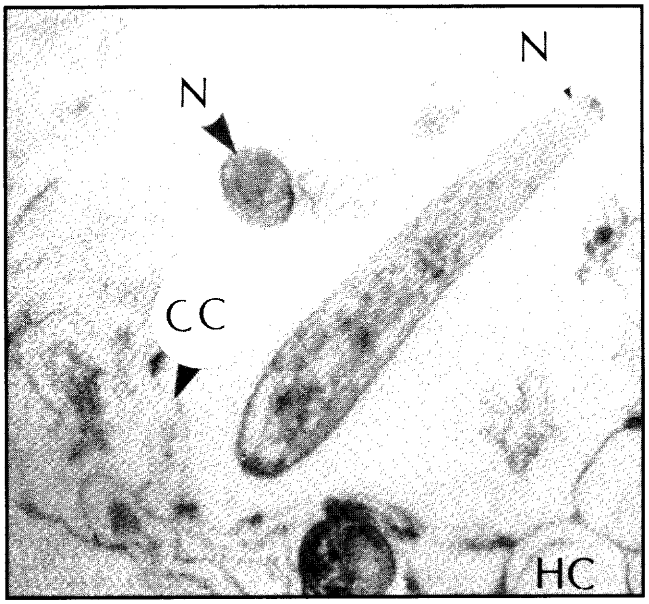

Fig. 3. Longitudinal section of the endocarp of the hull of a peanut pod infected by *Ditylenchus africanus*. Note cross and longitudinal sections of the nematode (N) bodies, collapsed cells (CC) and healthy cells (HC).

Ditylenchus africanus affects the yield of peanut seed by a) inducing second-generation seedlings, b) reducing fresh seed weight and c) decreasing seed quality by increasing the number of blemished and unsound seeds. The symptoms affecting seed quality are highly correlated with the number of nematodes in the testa (Venter *et al.* 1991). In greenhouse

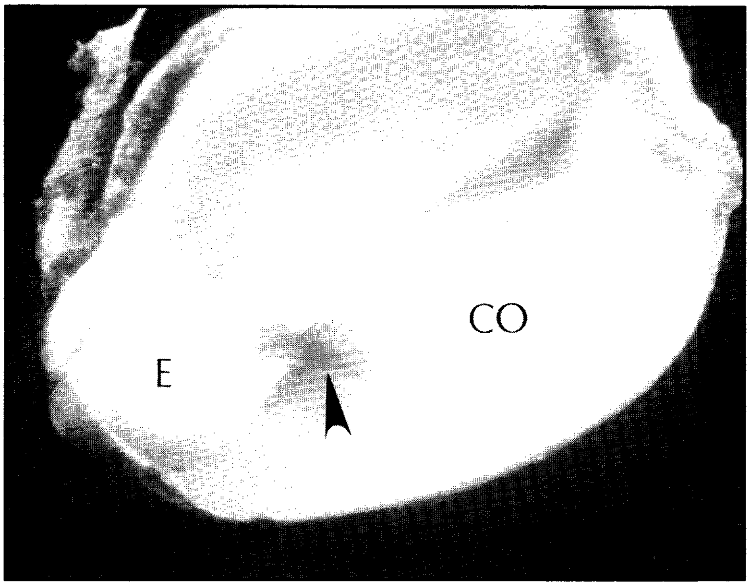

Fig. 4. Peanut seed infected by *Ditylenchus africanus*. The nematode does not infect the cotyledons (CO), but does infect the embryo (E), inducing necrosis (arrow).

experiments, an initial population density of 50 nematodes in the rhizosphere of a peanut seedling or a final nematode population of 20 nematodes per seed was sufficient to result in the downgrading of an edible seed consignment to undergrade seed (65 % or more blemished or unsound) (Venter *et al.* 1991).

SURVIVAL: *Ditylenchus africanus* can undergo dehydration and enter a state of anhydrobiosis. Either in anhydrobiosis or as eggs, the nematodes can survive in hulls left in the field in the absence of host plants for at least 32 weeks. This is long enough to survive the dry winter season in South Africa. With the spring rains the eggs hatch and the nematodes rehydrate (Basson *et al.* 1993). In whole seeds stored at 10° C (50° F), in cold storage for 24 weeks, relatively few nematodes survived, but the surviving nematode populations were high enough to build up large populations and to cause extensive damage (Basson *et al.* 1993).

MANAGEMENT: Since *D. africanus* can survive in stored seeds which may be symptomless, the use of nematode-free seeds is essential. Clean seeds can only be produced in nematode-free fields since treatment of infected seeds with nematicides or in a microwave oven was unsuccessful (unpublished results). Cleaning the soil of an already infested field is almost impossible because the nematodes can survive on hulls left in the field and on many crop plants, weeds and fungi. Removal of these hosts, can, however, help in keeping the nematode populations low.

In South Africa, non-volatile nematicides registered against *D. africanus* on peanut are phenamiphos (Nemacur®), oxamyl (Vydate®), and aldicarb (Temik®, Sanacarb®) at planting, and oxamyl and aldicarb at peg initiation. Nematicidal treatment usually gives economic control under irrigated production conditions, but does not give reliable control under South African dryland production conditions (McDonald and Van den Berg 1991). Since as few as 500 nematodes per plant at 12 weeks after planting can build up to damaging levels before harvest, any nematicide used should be active for longer than 12 weeks after planting (Basson *et al.* 1992). Soil fumigation with either methyl bromide or ethylene dibromide is the most efficient control measure against *D. africanus* in peanut fields (unpublished results).

Timely harvesting resulting in lower yield, but in the highest seed quality can give the best economic return (Basson *et al.* 1991; Venter *et al.* 1992)). The seeds should be harvested immediately after the appearance of the first symptoms on the pods. Since high numbers of eggs and anhydrobiotes occur with advanced ripening, timely harvest will also reduce the nematode densities in the hulls and seeds.

The peanut germplasm collection of Grain Crops Institute was screened for resistance to *D. africanus,* but of the more than 600 genotypes examined none showed complete resistance. A few genotypes had very low infections (Jordaan *et al.* 1989; Jordaan and van der Merwe 1989). Subsequently, a few more or less tolerant genotypes were identified (Van der Merwe and Joubert 1994; Venter *et al.* 1993). The most tolerant genotypes were from the Virginia bunch types which are characterized by a long growth cycle. The heritability for pod disease severity of the tolerant genotypes was relatively high, indicating possible avenues for genetic improvement via breeding for tolerance. Kwarts, a tolerant genotype, has been released for commercial use. However, its level of tolerance is inadequate under high nematode population pressure even when treated with a non-volatile nematicide (unpublished results).

Fields in the Northern Cape province of South Africa, in which peanuts have been grown in monoculture for many years, appear to be suppressive for the peanut pod nematode. From these fields, several nematode-trapping fungi have been isolated (Jones *et al.* 1996) including *Arthrobotrys* spp., *Monacrosporum* spp. and *Dactylaria* spp. The role of these fungi in the control of *D. africanus* is unknown.

SURVEY AND DETECTION: The outdoor climatic conditions of Florida are favorable to the establishment of *D. africanus* because the optimal soil temperature (28° C or 82° F) for nematode development and reproduction occurs during the peanut growing season (April-August) in the Florida peninsula. In case of accidental introduction of *D. africanus* into Florida, the nematode could become established and can survive on the weeds listed above. Examination of peanut pods is very important for the detection of this pest. Suspected necrotic pods and especially pods with corky appearance and dark tissues at junction with the peg should be collected from peanut fields and submitted to the Nematology laboratory to check for the presence of this nematode.

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